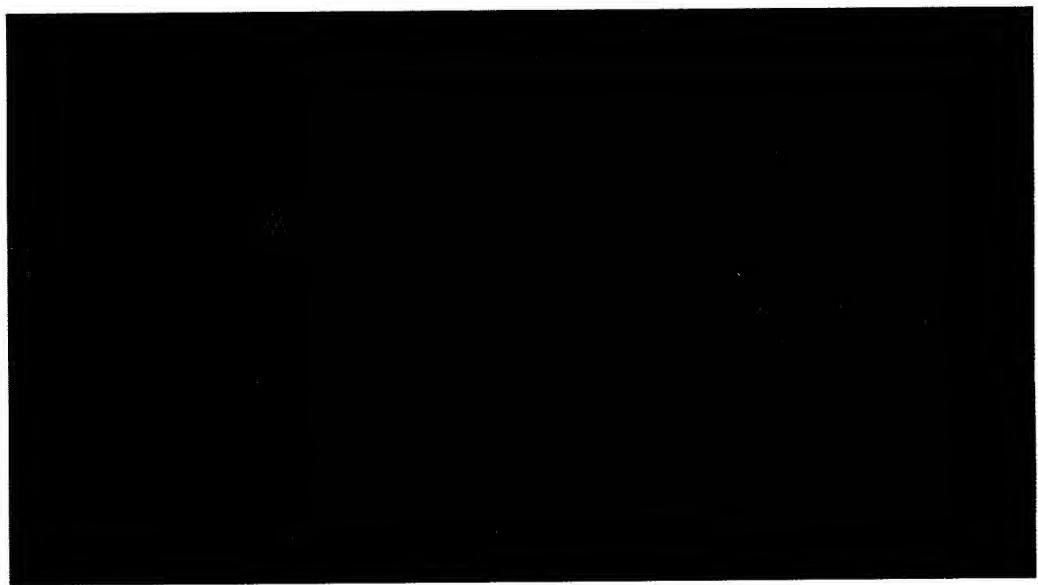


GFP-LCA (wt)

Figure 1



GFP-TLCA

Figure 2

1 PFVNKQENYK DPVNQVDIAY IKIPNVGQMQ PVKAFKIHNK IWVIPERDTE
51 TNPEEGDLNP PPEAKQVPVS YYDSTYLSTD NEKDNYLKGV TKLFERIYST
101 DLGRMILLTSI VRGIPFWGGS TIDTEALKVID TNCINVIQPD GSYRSEELNL
151 VIIGPSADII QFECKSFGHE VLNLTTRNGYG STQYIRFSPD FTFGFEESLE
201 VDTNPLLGAG KFATDPAVTL AHELIHAGHR LYGIAINPNR VEKVNTNAYY
251 EMSGLEVSFE ELRTFGGHDA KFIDSLQENE FRLYYY^{*}NKFK DIASTLINKAK
301 SIVGTTASLQ YMKNVFKEKY LLSEDTSGKF SVDKLKFDKL YKMLTEIYTE
351 DNFVKFFKVL NRKTYLNFDK AVEKINNIVPK VNYT^{*}YDGFN LRNTNLAANF
401 NGQNTEINNM NFTKLNKFTG ^{*}LFEFYKLLCV RGIITSK

Figure 3

GFP-LCA (AA mut)

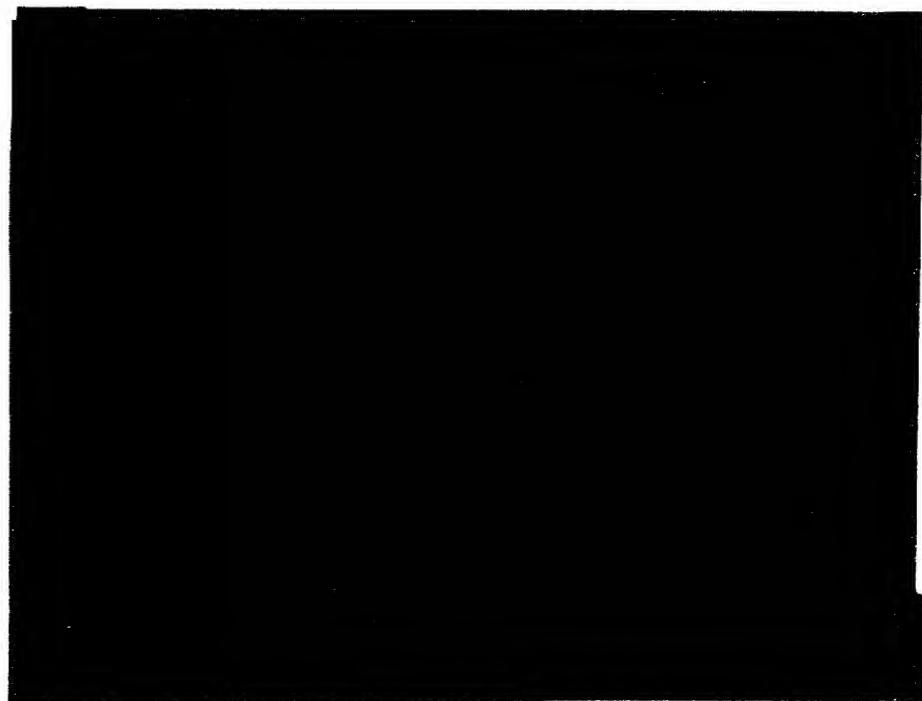


Figure 4

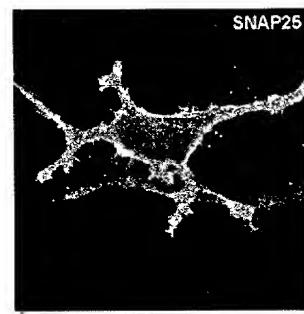


Figure 5

BOTULINUM TOXIN

TYPE A X-RAY STRUCTURE

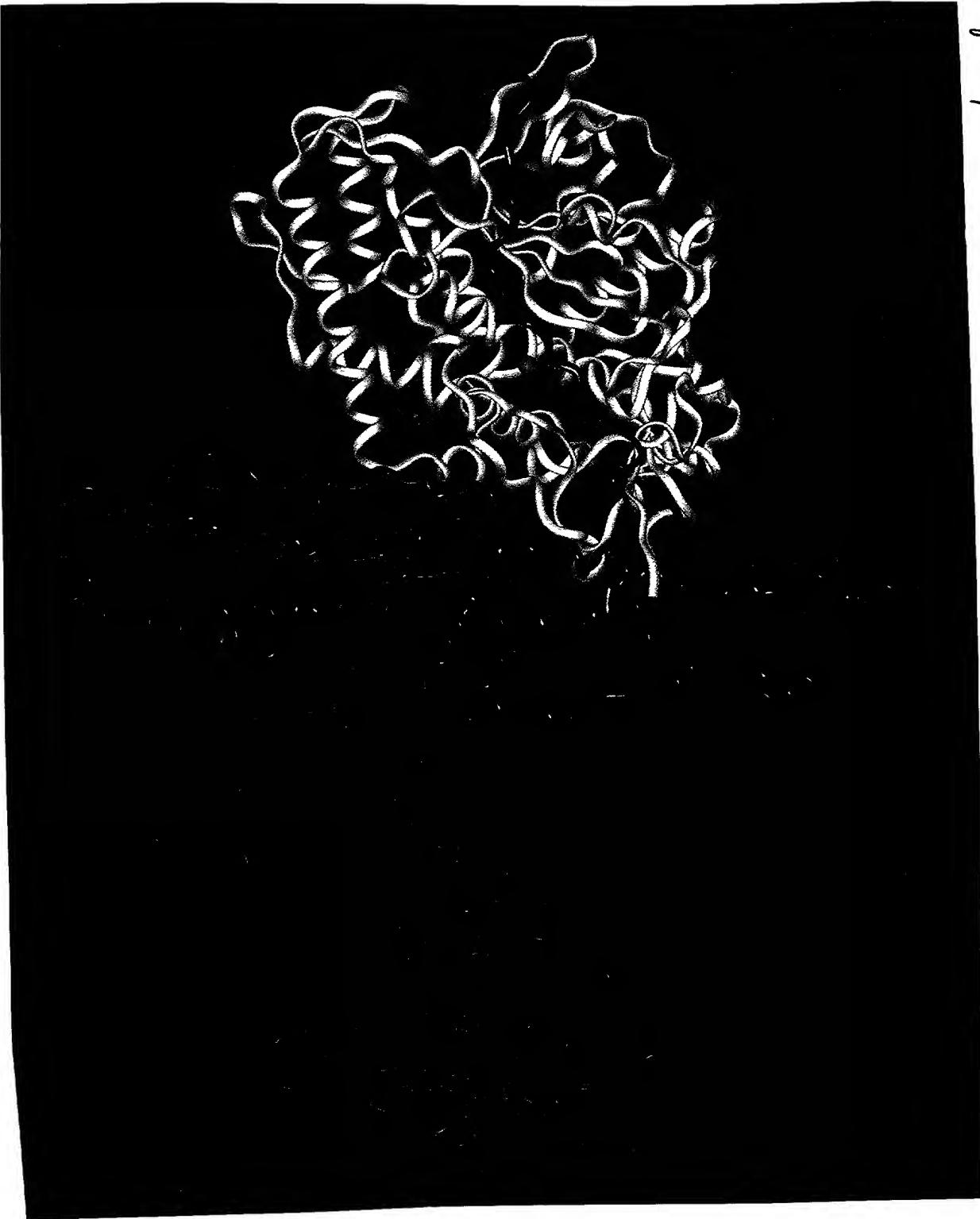
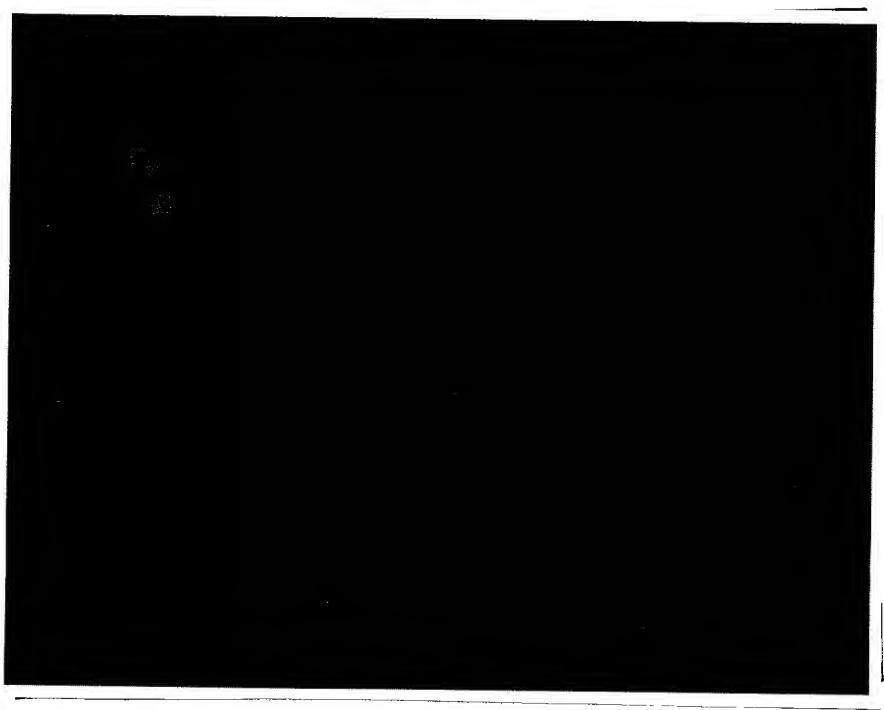


Figure 6

Green = binding domain
Red = + translocation domain
Yellow = protease domain



GFP-LCB(wt)

Figure 7

Figure 8

BONT-A_HallA_LC	(1)	- PFVNKQFENYQDFPVNGVDIAYIKIENVG-QMOPVIAFKI HINKIAWIPERDTHITWPE GDLNPPPEAKQWPVSYKID	75
BONT-B_Danish_I_LC	(1)	WVTRINNEMYNDIHDNNN IMMPEFARGTGRYKNAFKI DRDKI PERYTFGKPFDFNKSSGIFNRDVICEYID	
Consensus	(1)	P NENY DPI 1 1 P	YYD
BONT-A_HallA_LC	(74)	S7V1STDNEKDNYKGWTKLFERTYSTDTERMITSVRL1FVGSTEDTELKVIDTNCIN W1QPDGSYR-SEE	150
BONT-B_Danish_I_LC	(76)	PDYLNNDKUNIFQTMKLENRKGKPFKEKLEM INETIYUUDRYPUEFTNTIASV TANKLISNPGEVER	
Consensus	(76)	YL T K FL M KLF RI S LG LL II GIPF G 1 E 1 V	E
BONT-A_HallA_LC	(148)	151 --- LNIVLIGESADIIQFCKSP HEVNLNTIN STOYTR SDFTPCE EEESL WDTNPLLAGKFAHDDA	225
BONT-B_Danish_I_LC	(151)	KKGIPAMIIIFEGPVLNEN TDI QNHFASE EEGIMOMKTC EYVSVWNNVQENKGASTFNRGGYFS SDPA	
Consensus	(151)	NLII GP 1 E G SR GPG IKF PDF F E 1 P SDPA	
BONT-A_HallA_LC	(218)	226 VITLAELTHAGDRLYGIAINPNMRVFKV TNAVYEMSGLEVS FEELREFGCH MAXKF DSLOENEFRLYYVINKD	300
BONT-B_Danish_I_LC	(226)	LILMEEHTVLLGlyGKVD -DLPIVPEKKFFM QSTDATQAEEL OPPSIITPSTDKSIYDKVLQMDRG	
Consensus	(226)	L L HELH H LYGI I N FF S 1 EEL TFGG D 1 D N FK I	
BONT-A_HallA_LC	(293)	301 ASTMIAKASIVG-TTASLQYMIYFPERKYL LLSETSKRS WOKLKFLKIVWPLTB YEDIPVKFFVILN KW	375
BONT-B_Danish_I_LC	(300)	VDRHIVLVCISDPEININ YKFDYKVFV SEEKYS EVESFRKLN SNPGT FTMIAENY TKTNA MF	
Consensus	(301)	LNK I IN KN PKDY EDS GKFSD FDKLYK L FTE N FKI R SY	
BONT-A_HallA_LC	(367)	376 NFDKAVFKIN-IVPKVNTYD DETEFRNTNLAANFNEQTE INMMETKJKNFTGLF FEFYKILCVRGITLSK	447
BONT-B_Danish_I_LC	(375)	SDSLPPV KI KNWLDNEIYT EGEN ESDKNEKEYRE OMKAYBEIS-- KA HLAVK QMC SKV--	
Consensus	(376)	KI IL YTI DGFNI L F GQN IN F 1 YKI K I	

BirA-SNAP25 Assay of rLC/A Activity

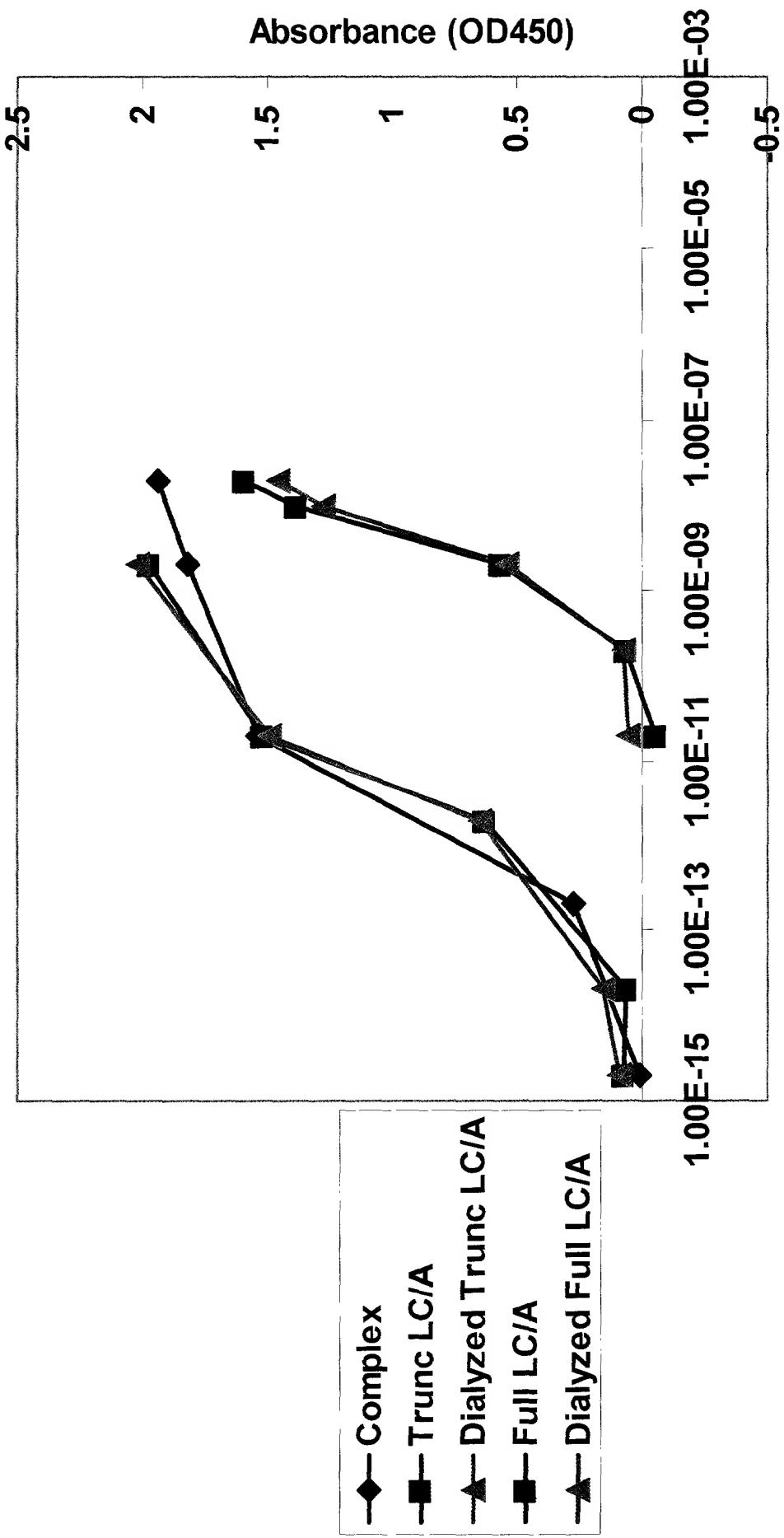


Figure 9

Comparison of LC/A constructs expressed from *E. coli* for *in vitro* analysis

Wildtype LC/A	...NFTKLKNFTGL EEFYYKLL CVRGIITSK	6xHis
trun LC/A	...NFTKL EEFYYKLL CVRGIITSK	S-tag
N-His LC/A	...NFTKLKNFTGL EEFYYKLL CVRGIITSK	6xHis

- Truncated LC construct published
 - Kadkhodayan, S. et al. *Prot. Exp. Purif.* 2000, 19, 125-130
 - Crystal structure reported at IBRCC in Oct. 2000

Figure 10